

IN THE U.S. PATENT AND TRADEMARK OFFICE

APPLICANT:

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FOR:

SIALOGOGUE, ORAL COMPOSITION AND FOOD PRODUCT  
CONTAINING THE SAME

D E C L A R A T I O N

Honorable Commissioner of Patents  
Washington, D.C. 20231

Sir,

I, Takashi Kojima, a patent attorney of Ginza  
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Japan do hereby solemnly and sincerely declare:

1) THAT I am well acquainted with Japanese language  
and English language;

2) THAT the attached is a full, true and faithful  
translation into English made by me of the PCT application  
of which number is PCT/JP2004/017328, filed in Japan  
on the 16 November 2004.

3) THAT I declare further that all statements made herein of my own knowledge are true and all statements made on information and belief are believed to be true, and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code and that such willful statements may jeopardize the validity of the application or any patent issued thereon.

AND I being sworn state that the facts set forth above are true.

Dated this *2nd* day of May 2006

  
Takashi KOJIMA

DESCRIPTIONSIALOGOGUE, ORAL COMPOSITION AND  
FOOD PRODUCT CONTAINING THE SAME

5

TECHNICAL FIELD

The present invention relates to a sialogogue for  
curing mouth dryness and an oral composition and food product  
10 incorporated therewith.

BACKGROUND ART

Xerostomia (dry mouth) is a physiological phenomenon  
which is experienced in daily life. It gives sticky  
15 displeasure, makes speaking difficult, and causes bad breath.  
In its pathological state, it changes the oral microbial  
flora, thereby creating a failure in oral functions such as  
dental caries, periodontal disease, and mucosal infectious  
disease. Consequently, promoting salivary secretion and  
20 thereby wetting the oral cavity are important in making the  
oral cavity feel refreshed and preventing oral diseases.

So, it has been recognized that wetting the oral  
cavity is necessary to make the oral cavity feel refreshed  
and prevent oral diseases. To meet this requirement, there  
25 has been proposed the use of hyaluronic acid (which is a  
humectant) in W000/56344. There has also been proposed an  
idea of positively promoting salivary secretion, thereby  
wetting the oral cavity. To materialize this idea, there has  
been proposed the use of a sialogogue selected from pickled  
30 Japanese apricot or Japanese apricot vinegar (in Japanese  
Patent Laid-Open No. Sho 56-22719) or organic acid (in  
Japanese Patent Laid-Open No. Hei 7-101856). There have also  
been proposed other sialogogues which do not resort to acid  
stimulation. Japanese Patent Laid-Open No. Hei 10-182392  
35 discloses Cola nuts (Sterculiaceae) and Japanese Patent  
Laid-Open No. 2002-265375 discloses Capparis masaikai

(Capparidaceae), *Capparis pterocarpa* Chun (Capparidaceae) and *Centella asiatica* (Umbelliferae).

Regrettably, hyaluronic acid produces the moisturizing effect but does not positively promote salivary secretion.

5 Moreover, the above-mentioned sialogogues are limited in their use because they taste somewhat.

#### DISCLOSURE OF INVENTION

It is an object of the present invention to provide a  
10 sialogogue and an oral composition and food product incorporated therewith. The sialogogue is tasteless and capable of moisture retention. The oral composition incorporated with the sialogogue includes toothpaste, mouthwash, artificial saliva, denture stabilizer, and  
15 solution for the oral care system with water supply and suction functions. The food product includes swallowing assistant, chewing gum, candy, drinks, and gummi.

In order to achieve the above-mentioned object, the present inventors carried out a series of researches, which  
20 led to the finding that polyglutamic acid and salts thereof promote salivary secretion and produce moisturizing effect and are almost tasteless and hence capable of incorporation into oral compositions and food products without the possibility of impairing their taste. The present invention  
25 is based on this finding.

Thus, the present invention is directed to a sialogogue including polyglutamic acid or a salt thereof and also to an oral composition and a food product both incorporated with the sialogogue.

30 The sialogogue according to the present invention moisturizes the oral mucous membrane of a patient suffering from serious dry mouth. Therefore, it easily solves problems with sticky displeasure, difficulties in speaking, and bad breath. In addition, it also prevents a failure in oral  
35 functions, such as dental caries, periodontal disease, and mucosal infectious disease. Being almost tasteless, it can be incorporated into oral compositions and food products

without restrictions. Moreover, being known as the sticking component of natto (fermented soybeans), it is highly safe and suitable for oral compositions and food products.

5                                    BRIEF DESCRIPTION OF DRAWINGS

Fig. 1 is a graph showing how the cumulative amount of saliva changes with time after administration of sodium  $\gamma$ -polyglutamate (with an average molecular weight of 1,200,000) in Experiment Example 1.

10                                  Fig. 2 is a graph showing how the cumulative amount of saliva changes with time after administration of potassium  $\gamma$ -polyglutamate (with an average molecular weight of 1,200,000) in Experiment Example 1.

Fig. 3 is a graph showing how the cumulative amount of saliva changes with time after administration of sodium  $\gamma$ -polyglutamate (with an average molecular weight of 300,000) in Experiment Example 1.

Fig. 4 is a graph showing how the cumulative amount of saliva changes with time after administration of sodium hyaluronate in Experiment Example 1.

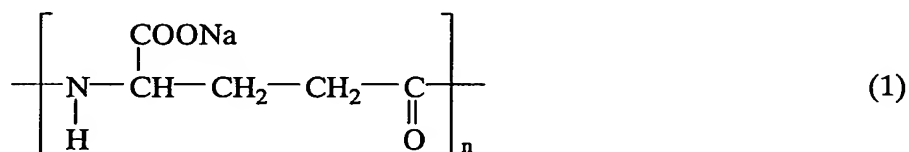
BEST MODE FOR CARRYING OUT THE INVENTION

The sialogogue according to the present invention is polyglutamic acid, which is chemically synthesized  $\alpha$ - or  $\gamma$ -polyglutamic acid or a salt thereof, or natural  $\alpha$ - or  $\gamma$ -polyglutamic acid or a salt thereof obtained from a variety of strains as a product of fermentation. Natural polyglutamic acid is desirable for incorporation into oral compositions and food products. Most desirable is  $\gamma$ -polyglutamic acid which is industrially available in large quantities. The polyglutamic acid may be either of D-form or L-form. The polyglutamic acid is insoluble in water but its salt is soluble in water. The salt includes sodium salt, potassium salt, magnesium salt, calcium salt, ammonium salt, ethanolamine salt, and basic amino acid salt. Any salt may

be used which is suitable for oral compositions and food products. The polyglutamic acid may be neutralized to an adequate degree which is known from the fact that a 1 wt% aqueous solution of the polyglutamate marks pH 1 to pH 14.

5 The polyglutamic acid used in the present invention is not specifically restricted in molecular weight. However, the weight average molecular weight (in terms of sodium salt measured by the method mentioned later) should be 10,000 to 5,000,000, preferably 20,000 to 4,000,000, more preferably 10 40,000 to 3,000,000, and most preferably 50,000 to 2,000,000. An adequate molecular weight should be selected according to the type of the product.

Sodium polyglutamate represented by the formula (1) below is particularly desirable.



15

(wherein n is an integer of 66 to 33,112, especially 331 to 13,245.)

The polyglutamic acid or a salt thereof to be used as the sialogogue according to the present invention should be 20 added to oral compositions or food products in an amount of 0.001 to 10 % by weight, preferably 0.005 to 7 % by weight, more preferably 0.01 to 5 % by weight, and most desirably 0.05 to 3 % by weight. An amount less than the lower limit will not produce the desired effect, and an amount more than 25 the upper limit will increase viscosity to adversely affect the feeling.

The sialogogue according to the present invention may be administered 1 to 6 times a day, with each dose ranging from 0.01 to 1 g, which is enough to promote salivary 30 secretion.

The sialogogue according to the present invention may be incorporated into oral compositions and food product. The

oral compositions include toothpaste, mouthwash, chewing tablet, oral ointment, gargling tablet, troches, artificial saliva, denture stabilizer, and solution for the oral care system with water supply and suction functions. The food product includes swallowing assistant, candy, chewing gum, drinks, and gummi. The sialogogue may be used in combination with any known component according to the kind and form of the oral composition and food product.

Those components used for oral compositions in the form of liquid or paste include humectant, binder, surfactant, sweetener, antiseptic, colorant, flavor, and other effective ingredients. They may be mixed with water to produce the desired products. Another component used for toothpaste is an abrasive.

The humectant is exemplified by polyhydric alcohols such as sorbitol, glycerin, propylene glycol, polyethylene glycol, xylitol, multitol, and lactitol. Its amount should be 5 to 50 % by weight, particularly 20 to 45 % by weight for paste products, and 0 to 50 % by weight, particularly 1 to 20 % by weight for liquid products.

The binder is exemplified by carrageenan, sodium hydroxyethyl cellulose, sodium carboxymethyl cellulose, hydroxyethyl cellulose, hydroxypropyl cellulose, methyl cellulose, hydroxypropylmethyl cellulose, sodium alginate, propylene glycol ester of alginic acid, polyacrylic acid, sodium polyacrylate, xanthan gum, talha gum, guar gum, locust bean gum, jellan gum, gelatin, curdlan, gum Arabic, agar, pectin, polyvinyl alcohol, polyvinyl pyrrolidone, and pullulan. The amount of the binder ranges from 0.1 to 5 % by weight for paste products (such as toothpaste) and 0 to 5 % by weight for liquid products (such as liquid dentifrice and mouthwash).

The surfactant is exemplified by anionic surfactant, cationic surfactant, and nonionic surfactant. Their typical examples include sodium lauryl sulfate, sodium  $\alpha$ -olefinsulfonate, N-acylglutamate, 2-alkyl-N-carboxylmethyl-N-hydroxyethylimidazolinium betaine, N-acyltaurate, sugar

fatty acid ester, alkylolamide, polyoxyethylene hardened  
castor oil, polyglycerin fatty acid ester,  
polyoxyethylene-polyoxypropylene glycol,  
polyoxyethylenesorbitan monostearate, lauroylsarcosine sodium,  
5 alkylpolyglucoside, and polyoxyethylene alkyl ether  
sulfosuccinate. The amount of the surfactant is usually 0.5  
to 5 % by weight.

The sweetener is exemplified by saccharine sodium,  
stevioside, stevia extract, paramethoxycinnamic aldehyde,  
10 neohesperidyl hydrochalcone, and perillartine. The colorant  
is exemplified by Blue No. 1, Yellow No. 4, and titanium  
dioxide. The antiseptic is exemplified by  
parahydroxybenzoate ester and sodium benzoate.

The flavor is exemplified by terpenes and their  
15 derivatives (such as 1-menthol, carvone, anethole, and  
limonene) and peppermint oil.

The liquid agents may be incorporated with a nontoxic  
solvent (such as ethanol) in an amount of 0 to 30 % by weight,  
particularly 1 to 25 % by weight.

20 The abrasive is exemplified by silica gel,  
precipitated silica, aluminosilicate, zirconsilicate,  
dibasic calcium phosphate anhydride or dihydrate, calcium  
pyrophosphate, calcium tertiary phosphate, hydroxyapatite,  
calcium carbonate, aluminum hydroxide, alumina, magnesium  
25 carbonate, magnesium tertiary phosphate, zeolite, zirconium  
silicate, and plastic-based abrasive.

The preferred amount of the abrasive ranges from 10 to  
50 % by weight for toothpaste and 0 to 30 % by weight for  
liquid dentifrice.

30 The artificial saliva may be incorporated with  
potassium chloride (0.1 to 0.2 % by weight), sodium chloride  
(0.05 to 0.1 % by weight), calcium chloride (0.01 to 0.02 %  
by weight), and magnesium chloride (0.004 to 0.006 % by  
weight), and optional dipotassium hydrogenphosphate (0.01 to  
35 0.05 % by weight).

To prepare the oral compositions in solid form (such  
as tablets and troches), the sialogogue may be combined with



nontoxic filler, binder, disintegrator, lubricant, surfactant, sweetener, and flavor (the last three mentioned above).

The filler is exemplified by cellulose and its derivatives, starch and its derivatives, sugars, and sugar alcohol. Their specific examples include crystalline cellulose, lactose, white soft sugar, mannitol, corn starch, potato starch, hydroxypropylstarch, calcium silicate, calcium hydrogenephosphate anhydride, and magnesium aluminometasilicate.

The binder is exemplified by hydroxypropyl cellulose, hydroxypropylmethyl cellulose, methyl cellulose, polyvinyl alcohol, polyvinylpyrrolidone, gelatin, dextrin, starch, and alpha starch.

The disintegrator is exemplified by carmellose, carmellose calcium, croscarmellose sodium, low substituted hydroxypropylcellulose, low substituted sodium carboxymethyl starch, and crospovidone.

The lubricant is exemplified by magnesium stearate, calcium stearate, sucrose ester of fatty acid, anhydrous silicic acid, light anhydrous silicic acid, and sodium stearyl fumarate.

The amount of the filler should be 1 to 10 % by weight, particularly 3 to 5 % by weight. The amount of the binder should be 0.1 to 1 % by weight, particularly 0.2 to 0.3 % by weight.

The food product may be formed from different materials according to its kind. For example, chewing gum may contain 10 to 50 % by weight of saccharide (such as sugar) and 50 to 90 % by weight of gum base, and candy may contain 35 to 40 % by weight of starch syrup and 60 to 65 % by weight of sugar.

#### EXAMPLES

The invention will be described below in more detail with reference to Experiment Examples, Working Examples, and Comparative Examples, which are not intended to restrict the scope thereof. In the following examples, "%" means "% by

weight" unless otherwise mentioned. Also, in the following examples, weight average molecular weight (Mw) is represented by the one which is measured by GPC method.

5 GPC Method

A sample (2 mg) of polyglutamic acid is dissolved in 2 mL of 0.1 mol/L phosphate buffer solution (approximately pH 7.0). Each sample (2 mg) of pullulan P-82, pullulan P-10, pullulan P-50, pullulan P-200, and pullulan P-1600 is  
10 dissolved in 2 mL of 0.1 mol/L phosphate buffer solution to give standard solutions. The sample solution and the standard solution (each 50  $\mu$ L) undergo GPC test, and the peak top molecular weight is obtained by using C-R7A·GPC program (product of SHIMADZU CORPORATION). The thus obtained value  
15 is regarded as the molecular weight.

<Conditions of analysis>

Detector: differential refractometer  
Precolumn: Shodex Asahipak GS-IG 7B (product of Showa  
20 Denko K.K), 7.6 mm ID  $\times$  100 mm (or equivalent)  
Main column: Shodex Asahipak GF-710 HQ (product of Showa Denko K.K), 7.6 mm ID  $\times$  300 mm + Shodex Asahipak GF-510 HQ (product of Showa Denko K.K), 7.6 mm ID  $\times$  300 mm (or equivalent)  
25 Column temperature: constant at about 40°C  
Mobile phase: 0.1 mol/L phosphate buffer solution  
(prepared by dissolving 7.1 g of disodium hydrogenphosphate anhydride ( $\text{Na}_2\text{HPO}_4$ ) and 6.8 g of potassium dihydrogenphosphate ( $\text{KH}_2\text{PO}_4$ )  
30 in water just enough to make one liter.)  
Flow rate: 0.5 mL/min  
Measurement time: 60 minutes.

### Experiment Example 1

To evaluate the effect of promoting salivary secretion

A sample of mouthwash was prepared from an aqueous solution containing 0.1% each of  $\gamma$ -polyglutamate and sodium  
5 hyaluronate. This sample was tested for the effect of promoting salivary secretion in the following manner.

Test for promotion of salivary secretion:

Three panelists, who were made to feel thirsty by  
10 heavy exercise in the previous day, were tested for saliva secretion by the following steps which followed one after another.

- (1) Mouth washing for 30 seconds with 20 mL of physiological saline.
- 15 (2) Spitting of saliva into a sputum mug, and measurement of the cumulative amount of saliva (for 30 minutes) at intervals of five minutes.
- (3) Mouth washing for 30 seconds with 20 mL of the sample solution.
- 20 (4) Measurement of the cumulative amount of saliva (for 30 minutes) in the same way as in (2).

Table 1 shows an increase (on average) from salivary secretion after mouth washing with physiological saline to  
25 salivary secretion after mouth washing with the sample solution. Figs. 1 to 4 show the cumulative amount of saliva measured at intervals of five minutes.

Table 1

Mouthwash solution	Ratio of increase in saliva secretion (%)
0.1% sodium $\gamma$ -polyglutamate (Mw = 1,200,000)	151
0.1% potassium $\gamma$ -polyglutamate (Mw = 1,200,000)	128
0.1% sodium $\gamma$ -polyglutamate (Mw = 300,000)	132
0.1% sodium hyaluronate (for comparison)	109

It is apparent from Table 1 and Figs. 1 to 4 that  
5  $\gamma$ -polyglutamic acid promotes salivary secretion.

Experiment Example 2

To evaluate the wet feeling and taste

Samples (0.1% aqueous solutions shown in Table 2) were  
10 prepared as shown in Table 2. Three panelists washed their mouth for 30 seconds with 20 mL of each sample solution and then spitted it out. Fifteen minutes later, the panelists rated the oral wet feeling and refreshed feeling according to the following criterion. Table 2 shows the results of rating  
15 (total points given by the three panelists).

Table 2

Samples	Wet feel	Taste	Feature of taste
Sodium $\gamma$ -polyglutamate (Mw = 1,200,000)	◎	○	Tasteless
Potassium $\gamma$ -polyglutamate (Mw = 1,200,000)	◎	○	Tasteless
Sodium $\gamma$ -polyglutamate (Mw = 300,000)	◎	○	Tasteless
Citric acid (for comparison)	×	×	Acidic
Cola nuts: water extract (for comparison)	×	△	Slightly spicy
Capparis masaiikai seeds: 50% ethanol extract (for comparison)	×	×	Bitter sweet
Sodium hyaluronate (for comparison)	○	○	Tasteless

\*The  $\gamma$ -polyglutamic acid is the same one as used in Experiment Example 1.

### Rating of wet feel

Standard for rating:

Degree	Strong	Weak	Slight	None
Point	3	2	1	0

Rating:

Total points	7 to 9	4 to 6	1 to 3	0
Degree	Strong	Weak	Slight	None
Rating	◎	○	△	×

5

### Rating of taste

Degree	None	Weak	Strong
Rating	○	△	×

10            Examples 1 to 18 that follow show the formulations.  
Incidentally, the polyglutamate has a degree of  
neutralization at pH 7. A good effect of promoting salivary  
secretion was produced by all the samples of oral  
compositions and food products.

15

#### [Example 1] Toothpaste

	Precipitated silica	25.00%
	Glycerin	25.00
20	Sorbit	15.00
	Xylitol	10.00
	Lauroyl decaglycerin ester	1.00
	Myristic acid diethanolamide	2.00
	Flavor	1.00
25	Saccharin sodium	0.20
	Calcium $\gamma$ -polyglutamate (Mw = 1,000,000)	0.10
	Purified water	balance
	Total	100.0%

[Example 2] Liquid dentifrice

	Aluminum hydroxide	25.00%
	Glycerin	40.00
	Sorbit	15.00
5	Carboxymethyl cellulose (DP = 500)	0.20
	Propylene glycol	2.00
	Sodium laurate	1.50
	Decaglyceryl monolaurate	1.00
	Flavor	1.00
10	Saccharin sodium	0.10
	Sodium $\gamma$ -polyglutamate (Mw = 1,500,000)	0.10
	Purified water	balance
	Total	100.0%

15 [Example 3] Oral ointment

	Liquid paraffin	15.00%
	Cetanol	10.00
	Glycerin	20.00
	Polyoxyethylene sorbitan fatty acid ester	5.00
20	Flavor	0.50
	Saccharin sodium	0.10
	Lysine $\gamma$ -polyglutamate (Mw = 300,000)	0.20
	Purified water	balance
	Total	100.0%

25

[Example 4] Mouthwash

	Ethanol	20.00%
	Flavor	1.00
	Polyoxyethylene (EO 60) hydrogenated castor oil	0.30
30	Sodium monofluorophosphate	0.10
	Saccharin sodium	0.05
	Sodium $\gamma$ -polyglutamate (Mw = 1,200,000)	0.20
	Purified water	balance
	Total	100.0%

[Example 5] Gargle tablet

	Sodium bicarbonate	53.0%
	Citric acid	18.0
	Anhydrous sodium sulfate	12.0
5	Dibasic sodium phosphate	10.0
	Polyethylene glycol	3.0
	Flavor	2.0
	<u>Ammonium <math>\gamma</math>-polyglutamate (Mw = 80,000)</u>	<u>2.0</u>
	Total	100.0%

10

[Example 6] Troche

	Xylitol	92.0%
	Gum acasia	5.0
15	Talc	2.0
	Magnesium stearate	0.7
	<u>Potassium <math>\gamma</math>-polyglutamate (Mw = 1,200,000)</u>	<u>0.3</u>
	Total	100.0%

20

[Example 7] Chewing tablet

	Erythritol	85.0%
	Potato starch	4.0
	Talc	3.5
25	Magnesium stearate	1.5
	Citric acid	5.0
	<u>Arginine <math>\gamma</math>-polyglutamate (Mw = 1,200,000)</u>	<u>1.0</u>
	Total	100.0%

[Example 8] Denture stabilizer (in gum form)

	Vinyl acetate resin	60.0%
	Light calcium carbonate	3.0
	Yellow beeswax	3.0
5	Polypropylene glycol	3.0
	Ethanolamine $\gamma$ -polyglutamate (Mw = 1,500,000)	1.0
	<u>60% ethanol</u>	<u>balance</u>
	Total	100.0%

10

[Example 9] Denture stabilizer (in powder form)

	Sodium carboxymethyl cellulose	74.0%
	Polyethylene oxide	24.0
	<u><math>\gamma</math>-polyglutamic acid (Mw = 300,000)</u>	<u>2.0</u>
15	Total	100.0%

[Example 10] Denture stabilizer (in paste form)

	Sodium carboxymethyl cellulose	32.0%
20	Polyethylene oxide	13.0
	Vaseline	40.0
	Arginine $\gamma$ -polyglutamate (Mw = 1,000,000)	1.0
	pH adjustor	0.2
	Flavor	0.1
25	Antiseptics	q.s.
	Coloring matter	q.s.
	<u>Liquid paraffin</u>	<u>balance</u>
	Total	100.0%



[Example 11] Mouth refrigerant

	Ethanol	30.0%
	Xylitol	10.0
	Flavor	2.0
5	Polyoxyethylene (EO 60) hydrogenated castor oil	1.5
	Potassium $\gamma$ -polyglutamate (Mw = 1,000,000)	1.0
	Purified water	balance
	Total	100.0%

10 [Example 12] Solution for the oral care system with  
water-supply and suction functions

	Glycerin	2.0%
	Xylitol	2.0
	Polyoxyethylene (EO 60) hydrogenated castor oil	1.0
15	Ethanolamine $\gamma$ -polyglutamate (Mw = 1,200,000)	0.5
	pH adjustor	0.2
	Flavor	0.2
	Antiseptics	q.s.
	Coloring matter	q.s.
20	Purified water	balance
	Total	100.0%

[Example 13] Artificial saliva

	Potassium chloride	0.15%
25	Sodium chloride	0.06
	Magnesium chloride	0.005
	Calcium chloride	0.015
	Hydroxypropyl cellulose	0.1
	Glycerin	1.0
30	Calcium $\gamma$ -polyglutamate (Mw = 1,500,000)	0.5
	Flavor	0.02
	Antiseptics	q.s.
	Purified water	balance
	Total	100.0%

[Example 14] Swallowing assistant (4.5 g for 100 mL water)

	Xanthan gum	20.0%
	Guar gum	4.0
	Ammonium $\gamma$ -polyglutamate (Mw = 300,000)	1.0
5	Dextrin	balance
	Total	100.0%

[Example 15] Candy

10	Sugar	50.0%
	Starch syrup	33.0
	Organic acid	2.0
	Flavor	0.2
	Sodium $\gamma$ -polyglutamate (Mw = 300,000)	0.1
15	Purified water	balance
	Total	100.0%

[Example 16] Chewing gum

20	Sugar	53.4%
	Gum base	20.0
	Glucose	10.0
	Starch syrup	16.0
	Flavor	0.5
25	$\gamma$ -polyglutamic acid (Mw = 80,000)	0.1
	Total	100.0%

[Example 17] Drinks

	Glucose	1.35%
	Fructose	1.35
	Milk component	0.1
5	Sodium $\gamma$ -polyglutamate (Mw = 80,000)	0.1
	Sodium chloride	0.029
	Vitamin C	0.03
	Vitamin B1	0.00022
	Valine	0.0384
10	Leucine	0.0462
	Isoleucine	0.0154
	Citric acid	0.01
	Malic acid	0.01
	Flavor	0.01
15	Water	balance
	Total	100.0%

[Example 18] Gummi

20	Sugar	40.2%
	Starch syrup	48.2
	Gelatin	8.0
	Fruit juice	2.0
	Citric acid	0.5
25	Malic acid	0.5
	Flavor	0.5
	Calcium $\gamma$ -polyglutamate (Mw = 300,000)	0.1
	Total	100.0%